

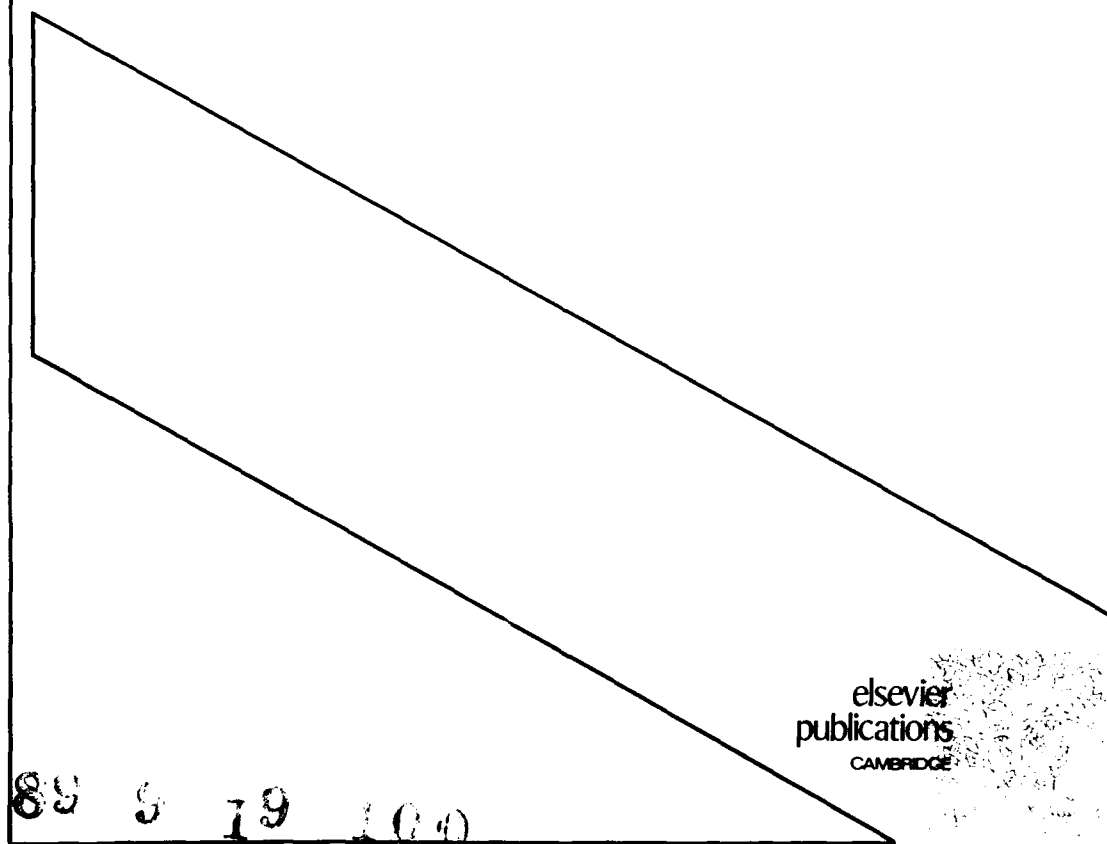
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Endogenous opioid peptides and epilepsy: quieting the seizing brain?

Frank C. Tortella

The brain opioid system has been implicated in the pathophysiology of seizure disorders and cellular mechanisms of epileptogenesis. While opioid peptides were originally envisioned as endogenous convulsants, a growing body of evidence demonstrates that these neuropeptides are also anticonvulsant. A role for opioid peptides as neuromodulators of postictal seizure arrest and refractoriness is recognized, and their endogenous activation by seizures is firmly established. Frank Tortella reviews the proconvulsant/anticonvulsant pharmacology of opioid peptides, the evidence for their involvement in postictal mechanisms, and the possible existence in the CNS of an endogenous anticonvulsant substance with opioid peptide-like characteristics.

Clinical and experimental reports describing proconvulsant and anticonvulsant properties of opium date back to the 19th century¹. Over 150 years later this paradoxical relationship between opioids and seizures persists, nurtured over the past decade by studies aimed at determining the possible role of endogenous opioid peptides in epilepsy.

The idea that opioid peptides

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were endogenous convulsants was based solely on early EEG studies in rats demonstrating that large doses of enkephalin or β -endorphin caused nonconvulsive epileptic discharges^{2,3}. Unfortunately, simple demonstration of paroxysmal activity provides insufficient evidence for epileptogenesis. For example, some very potent GABA-mimetics, remarkable anticonvulsants themselves, can under certain conditions induce paradoxical EEG epileptiform activity⁴. The idea of opioid peptides being epileptogenic may therefore have been premature, and indeed it was

soon discovered that exogenously administered opioid peptides suppress, rather than potentiate, experimentally induced convulsions⁵.

Hence, in terms of their possible role in epilepsy, a complex and at times seemingly controversial profile has emerged. However, in attempting to elucidate the cellular mechanisms of epileptogenesis and the modulation of postictal seizure arrest and refractoriness, the question of how opioid peptides may be involved in the pathophysiology of epilepsy takes on considerable significance. This review focuses on the behavioral, electrophysiological and anatomical evidence for a role of endogenous opioid peptides in epilepsy and attempts to address the conflicting results and interpretations which have emerged regarding the proconvulsant/anticonvulsant paradox.

Proconvulsant effects

Shortly after the discovery of enkephalins and β -endorphin, one of the first observations made was that central injections of these opioid peptides in rats resulted in brief, transient bursts of epileptiform EEG activity^{2,6-8}. Convulsive motor activity was not associated with the epileptiform EEG, although under certain experimental conditions myoclonic and/or

TABLE I. Behavioral proconvulsant effects of opioid peptides

Experimental model (rats)	Ligand(s) tested	Route of administration	Sensitivity to opioid antagonist	Ref.
Metrazol test	β -endorphin, α -NEO, DADL	i.c.v.	yes (naloxone) for DADL only	a
Spontaneous convulsion:				
sialogic	β -endorphin	i.c.v.	n.t.	b
myoclonic	DAME, DADL, morphiceptin, DSTLE	i.c.v. intrahippocampal, i.c.v.	n.t. n.t.	c d
kindled	β -endorphin, [Met]enkephalin	site specific	yes (naloxone)	e
myoclonic	DADL	intrathalamic	yes (naloxone)	f
generalized	PLO17, DAMGO, DADL	intrahippocampal	yes (β -fNA)	g,h

*Prezwlocka, B. *et al.* (1983) *Life Sci.* 33, 595-598; *Holaday, J. W. *et al.* (1978) *Life Sci.* 22, 1525-1536; *Dzolic, M. R. and Poel-Heisterkamp, A. L. (1982) *Brain Res. Bull.* 8, 1-6; *Haffmans, J. and Dzolic, M. R. (1983) *J. Neurol. Transm.* 57, 1-11; *Cain, D. P. and Corcoran, M. E. (1985) *Brain Res.* 338, 327-336; *Walker, G. E. and Yaksh, T. L. (1986) *Brain Res.* 383, 1-14; *Lee, P. H. K. *et al.* (1988) *Brain Res.* 441, 381-385; *Lee, P. H. K. *et al.* *J. Neurosci.* (in press)
n.t., not tested

masticatory jaw movements have been observed^{9,10}. Usually, the ictal discharges are associated with behavioral immobilization interrupted only by intermittent bursts of wet-shake behavior. Subcortical EEG analysis of the seizure activity revealed a focus originating primarily in hippocampal/limbic structures, rapidly and synchronously generalizing to the cerebral hemispheres^{6,11,12}. Subsequent autoradiographic studies of cerebral metabolism targeted the limbic forebrain, hippocampus, lateral septum and amygdaloid nucleus as the primary focus of opioid peptide epileptiform activity^{12,13}. Interestingly, neuropathological alterations do not appear to be associated with the enkephalin EEG seizure¹⁰.

To a great extent, the electrophysiological and receptor mechanisms mediating this activity have been ascertained. The preponderant effect of opioid peptides on unit activity is inhibitory, hyperpolarizing neural elements throughout the CNS. However, intrinsic to the hippocampus, opioid peptides induce naloxone-reversible, single-unit excitations resulting from presynaptic disinhibition of inhibitory pyramidal interneurons. The end result of this disinhibition is excitation of pyramidal cell activity in the hippocampus, a brain area with discrete localization of opioid peptide neuronal circuitry and highly sensitive to epileptogenesis (see Refs 12 and 14). While direct enhancement of excitatory neurotransmission cannot be entirely ruled out, results of *in-vitro* and *in-vivo* receptor studies support the disinhibition mechanism of epileptiform activity. Both effects, i.e. disinhibition of hippocampal unit

activity and the resultant epileptiform EEG, are primarily mediated by specific interactions with μ -opioid receptors¹⁵⁻¹⁷ (although recent evidence suggests that δ -opioid receptors may also be involved^{18,19}).

Interestingly, while the major opioid-containing pathway in the hippocampus contains more prodynorphin-derived than pro-enkephalin-derived peptides¹⁴, dynorphin effects on CA3 neurons are mixed, exhibiting predominant inhibitory activity compared to enkephalin¹⁴. It is not surprising then that i.c.v. injections of dynorphin A fail to induce ictal EEG activity or behavioral convulsions^{11,20}. Thus, a major contribution of endogenous κ -opioid systems in the seizurogenic or convulsant actions of opioid peptides appears unlikely.

While this evidence provided a strong basis for the hypothesis that opioid peptides may be involved in the etiology of epilepsy and the cellular mechanisms of epileptogenesis, the EEG epileptiform activity observed in rats has not generalized to other species, and there is little evidence that exogenously administered, pharmacological doses of opioid peptides induce motor convulsions or increase seizure susceptibility in experimental models of epilepsy (Table I). Thus, defining proconvulsant activity based entirely upon EEG findings could be misleading.

While a nonspecific proconvulsant opioid system has been proposed³, there is now some evidence supporting a receptor-specific proconvulsant action of opioid peptides. It has been possible to kindle seizures with repeated injections of β -endorphin

which, in turn, can transfer to electrical kindling. The importance of this observation is as yet unclear since primary kindling sites do not have to be excited for transfer to occur. In addition, since moderate to high (1-10 mg kg⁻¹) doses of naloxone were required to partially limit β -endorphin kindling (Table I), this effect may not be entirely selective for opioid receptors. Nonetheless, other experiments using low doses of highly selective μ -opioid ligands have shown that generalized convulsions can be produced in rats following their single injections into the ventral hippocampus (Table I). More importantly, these convulsions could be prevented by pretreatment with the selective irreversible μ -opioid receptor antagonist β -funaltrexamine (β -fNA). It would appear, then, that under well defined conditions opioid peptides are capable of inducing convulsant activity, at least in rodents.

From a pathophysiological point of view the importance of these findings to the mechanisms of epileptogenesis remains ambiguous. Whether, like the nonconvulsive epileptiform EEG, these proconvulsant behaviors are also an artifact of the potent disinhibitory actions of opioid peptides in the hippocampus, or represent direct excitatory responses on neural elements reminiscent of other excitatory neurotransmitter systems, remains to be elucidated. The true importance of their convulsant properties in advancing our understanding of epilepsy may reside in their potential as developmental tools. For example, specific anti-petmal drugs have been shown to quiet the epileptiform EEG produced by large doses of enkephalin¹⁸,

TABLE II. Anticonvulsant effects of opioid peptides

Experimental model	Ligand(s)	Route of administration	Sensitivity to opioid antagonist	Ref.
MES test (mouse)	β -endorphin, DADL	i.c.v.	yes (naltrexone)	a
(rat)	β -endorphin, DAME, DADL	i.c.v.	yes (naloxone)	b
(rat)	DAME, dynorphin(1-13)	intranasal	yes (naloxone)	c
(rat)	DAMGO	i.c.v.	yes (naloxone, β -fNA)	d
(rat)	DPDPE	i.c.v.	yes (ICI-174864)	d
(rat)	DAMGO	i.c.v.	yes (CTP)	e
Kindling (rat)	ENK146104	i.p.	yes (naloxone)	f
(rat)	dynorphin(1-13)	intranasal	no (naloxone)	g
(rat)	DAMGO	i.v.	n.t.	h
Flurothyl (rat)	β -endorphin, DAME, DADL	i.c.v.	yes (naloxone)	i
(rat)	metkephamid, FK33-824	s.c.	yes (naloxone)	j
(rat)	DADL, metkephamid	i.c.v. or s.c.	yes (ICI-154129)	k
(rat)	DADL	i.c.v.	yes (naloxone, ICI-154129)	l
(rat)	dynorphin(1-13)	i.c.v.	no (naloxone)	m
(rat)	DAMGO	i.c.v.	yes (naloxone, β -fNA)	d
(rat)	DPDPE	i.c.v.	yes (ICI-154129)	d
Metrazol (rat)	DADL, dynorphin, α -NEO	i.c.v., intrathecal	yes (naloxone)	n
(rat)	[Leu]enkephalin	i.p.	no (naloxone)	o
Focal penicillin (rabbit)	[Met]enkephalin	intracortical	n.t.	p
(rabbit)	[Met]enkephalin	intrahippocampal	n.t.	q
Audiogenic (mouse)	[Met]enkephalin	i.v.	n.t.	p
(mouse)	[Met]enkephalin	i.c.v.	n.t.	r
(rat)	DAME	i.c.v.	n.t.	s
(rat)	[Leu]enkephalin, DAME, DADL, E ₂₅	i.c.v.	yes (ICI-174864); no (naloxone)	t
Photosensitive (<i>Papio papio</i>)	FK33-824	i.v.	yes (naloxone)	u
Epilepsy-prone (gerbil)	β -endorphin	i.c.v.	yes (naloxone)	v

*Puglisi-Allegr. S. et al. (1984) *Pharmacol. Biochem. Behav.* 20, 767-769; *Berman, E. F. and Adler, M. W. (1984) *Neuropharmacology* 23, 367-371; *Garant, D. S. and Gale, K. (1985) *J. Pharmacol. Exp. Ther.* 234, 45-48; *Tortella, F. C. et al. *Peptides* (in press); *Tortella, F. C. et al. (1988) *Proceedings of the International Narcotics Conference, Albi, France*; *Caldecott-Hazard, S. et al. (1982) *Brain Res.* 251, 327-333; *Bonhaus, G. A. et al. (1987) *Brain Res.* 405, 358-363; *Bohme, et al. (1987) *Neurosci. Lett.* 74, 227-231; *Tortella, F. C. et al. (1981) *Life Sci.* 29, 1039-1045; *Cowan, A. et al. (1981) *Eur. J. Pharmacol.* 71, 117-121; *Tortella, F. C. et al. (1983) *Life Sci.* 33, 603-606; *Tortella, F. C. et al. (1985) *Life Sci.* 37, 497-503; *Tortella, F. C. and Holaday, J. W. (1986) *NIDA Res. Monogr. Ser.* 74, 539-542; *Przewlocka, B. et al. (1983) *Life Sci.* 33, 595-598; *Costa de Costa, X. et al. (1987) *Epilepsia* 28, 584; *Baccelliere, L. et al. (1980) *Acta Neurol. Scand.* 62, 79; *Gusel, V. A. et al. (1985) *Farmakol. Toksikol. (Moscow)* 48, 28-31; *Plotnikoff, N. P. et al. (1976) *Life Sci.* 19, 1283-1288; *Kotlinska, J. and Langwinski, R. (1985) *Pol. J. Pharmacol. Pharm.* 37, 103-111; *Kotlinska, J. and Langwinski, R. (1986) *Drug Alcohol Depend.* 18, 361-367; *Meldrum, B. S. et al. (1979) *Brain Res.* 170, 333-348; *Bajorek, J. G. and Lomax, P. (1982) *Peptides* 3, 83-86
n.t., not tested

thereby providing a novel approach to study petit mal epilepsy and drug design.

Various opioid antagonists have been used in attempts to implicate endogenous opioid peptides as a causative influence in epilepsy. Indeed, very high doses of naloxone can induce generalized convulsions in rats (50 mg kg⁻¹, i.v.)^{12,21} or lower their convulsive threshold to flurothyl (200-300 μ g, i.c.v.) (FCT, unpublished). It appears however that these convulsant effects result from GABA antagonist properties of naloxone²¹. Experimentally, efforts to block endogenous 'convulsant' actions of opioid peptides with low doses of antagonists in various models of experimental seizures, including spontaneous seizures of a genetic origin, have failed.

In kindling, where a permanent change in seizure susceptibility exists in the absence of any genetic predisposition, the results with naloxone have also been conflict-

ing. Naloxone has been reported to have no effect, decrease kindling time and increase seizure severity, or have anticonvulsant activity (see Refs 3 and 22).

Clinical studies with naloxone, while being largely negative, have also been inconclusive²³. No direct anticonvulsant or proconvulsant effects of naloxone on clinical seizures have been defined, although in a small group of patients it is possible that some positive effects were observed²³. For the moment, however, the most plausible interpretation of the opioid antagonist studies is that endogenous opioid systems may not be critically important to the initiation and propagation of ictal events, or to the maintenance of seizure spread.

Anticonvulsant effects

In contrast to the proconvulsant effects of opioid peptides, their ability to suppress seizure activity in a dose-related manner, across

species, and in a variety of experimental models has been firmly established (Table II). Two independent observations predicted the anticonvulsant pharmacology of these peptides. Firstly, in the late 1970s it was reported that many, but not all, non-peptide opioids were anticonvulsant in rats²⁴. Secondly, detailed EEG studies revealed that in addition to the transient epileptiform activity opioid peptides induced sustained, high amplitude slow-wave patterns (EEG slowing) associated with behavioral depression, rather than excitation^{6-8,25}. Since our findings in 1981 demonstrating that enkephalin or β -endorphin raises seizure threshold in rats in a naloxone-reversible manner, anticonvulsant actions for opioid peptides have been defined in chemical, electrical and genetic models of epilepsy in mice, rats, gerbils, rabbits and baboons (Table II). With few exceptions (see below) these anticonvulsant effects can be

TABLE III. Anticonvulsant receptor profile in rat flurothyl test

Ligand tested	Antagonist			Receptor subtype	Ref.
	non-selective naloxone	μ -selective (irreversible) β -fNA	δ -selective ICI-154129/174864		
DAMGO	+++	+++	-*	μ	a
β -Endorphin	+	n.t.	n.t.	$\mu, \delta, \epsilon(?)$	b
Metkephamid	+	n.t.	+	μ and δ	c
DADL	+	-	+++*	μ and δ	b,d
DPDPE	n.t.	-	+++†	δ	a
Dynorphin(1-13)	-	n.t.	n.t.	non-opioid	e
Dynorphin(3-13)	-	n.t.	n.t.	non-opioid	e

Antagonist sensitivity pronounced (+++), moderate (+), no effect (-), not tested (n.t.) *ICI-154129; †ICI-174864

*Tortella, F. C. et al. *Peptides* (in press); †Tortella, F. C. et al. (1981) *Life Sci.* 29, 1039-1045; ‡Tortella, F. C. et al. (1983) *Life Sci.* 33, 603-606;

§Tortella, F. C. et al. (1985) *Life Sci.* 37, 497-503; ¶Tortella, F. C. and Holaday, J. W. (1986) *NIDA Res. Monogr. Ser.* 75, 533-542

blocked by opioid antagonists, indicating a specific, opioid receptor-mediated pharmacology.

In recent years we have turned our attention towards determining the opioid receptor subtype(s) responsible for this anticonvulsant pharmacology. Using a series of opioid peptides and antagonists with varied degrees of selectivity for μ -, δ - and κ -opioid receptors, an *in-vivo* receptor profile for anticonvulsant activity has been established⁵. In both the flurothyl test (a 'threshold' seizure model) (Table III) and the maximal electroshock (MES) test (a 'spreading' seizure model) (Table IV) opioid peptide-induced seizure protection can be selectively mediated by μ - and δ -receptors.

Here again the case of dynorphin is interesting. Dynorphin was first described as an anticonvulsant opioid in the rat flurothyl test (Table III). However, these effects were not antagonized with doses of naloxone as high as 10 mg kg⁻¹. Moreover, the opioid inactive fragment dynorphin A(3-13) was similarly anticonvulsant. Non-opioid anticonvulsant properties of dynorphin against kindled seizures have also been described (Table II). It appears in the case of dynorphin the paradox is that while not possessing EEG seizurogenic properties itself (unlike other opioids), its anticonvulsant effects involve non-opioid mechanisms. Although the anticonvulsant effects of

dynorphin are due in part to non-opioid mechanisms this should not be construed as a lack of importance for endogenous κ systems in seizure mechanisms. Indeed, the most selective κ -opioid U-50488, albeit not a peptide, exhibits direct inhibitory actions on CA3 neurons^{12,14} and is a potent anticonvulsant whose effects are antagonized by naloxone or the selective κ -antagonist nor-BNI (Table IV).

In general, the anticonvulsant effects of opioid peptides reflect their potent hyperpolarizing and cortical EEG slowing properties in the brain. It seems possible that these general inhibitory properties on CNS neurons could functionally lead to the suppression of seizure initiation, propagation or spread. Furthermore, it has been established that the hyperpolarizing^{15,16}, EEG slowing²⁵ and behavioral anticonvulsant properties⁵ represent specific pharmacological interactions with opioid receptors rather than high-dose, nonspecific toxicological effects. Therefore, the primary effect of low doses of exogenously administered opioid peptides appears to be seizure protection, with a pharmacological profile as anticonvulsants.

Seizure activation and post-seizure inhibition

An important observation associated with all the studies des-

cribing opioid peptide EEG and anticonvulsant activity (Table II) has been the failure of control injections of low (pharmacological) doses of opioid antagonists to effect spontaneous EEG, seizure threshold or spread. As a result, it is unlikely that endogenous opioid systems, be they proconvulsant or anticonvulsant, are tonically active.

There is, however, little doubt that seizures can 'turn on' endogenous opioids; some evidence indicates that peptides are released at higher firing rates, i.e. epileptic neurons, and that with higher frequencies there may be an increased functional effect of the peptide²⁶. Numerous preclinical studies have described an activation of opioid-like phenomena, including behavioral, EEG, biochemical and receptor changes, as a consequence of seizures^{5,27,28}. In the past two years alone, 15 preclinical reports have described increases in levels, gene expression, mRNA content or opioid receptor numbers following seizures. These findings are supported by recent clinical studies where increases in serum or CSF content of [Met]enkephalin or β -endorphin have been measured consequent to ECT, febrile or generalized tonic-clonic convulsions²⁹⁻³².

Since opioid peptides are anticonvulsant and activated by seizures it was proposed that they may function postictally to spontaneously arrest seizures and

TABLE IV. Anticonvulsant receptor profile in rat maximal electroshock test

Ligand tested	Antagonist					Receptor subtype	Ref.
	non-selective naloxone	μ -selective (irreversible) β -fNA	δ -selective ICI-154129/174864	κ -selective nor-BNI	μ -selective CTP		
DAMGO	+++	+++	-*	-	+++	μ	a,b
DPDPE	n.t.	-	+++†	-	-	δ	a,b
U50,488	+	n.t.	n.t.	+++	-	κ	b,c,d

Antagonist sensitivity pronounced (+++), moderate (+), no effect (-), not tested (n.t.) *ICI-154129; †ICI-174864

*Tortella, F. G. et al. *Peptides* (in press); †Tortella, F. C. et al. (1988) *Proceedings of the International Narcotics Research Conference, Albi, France*; ‡Tortella, F. C. et al. (1985) *J. Pharmacol. Exp. Ther.* 237, 49-53; §Tortella, F. C. et al. (1987) *Soc. Neurosci. Abstr.* 13, 208

Opioid peptide-like endogenous anticonvulsant substance

To directly address whether an endogenous opioid anticonvulsant substance (EAS) is present in the CNS following a seizure, we undertook an experimental approach similar to that employed by Otto Loewi over 60 years ago. We decided to remove cerebrospinal fluid (CSF) from donor rats during the postictal refractory period following a seizure (donor-CSF) and re-administer this donor-CSF directly into the brains of recipient animals. If the donor-CSF contained an endogenously activated anticonvulsant substance, then seizure protection should occur in these recipients when exposed to a subsequent convulsant. To avoid using a chemical toxin which in itself may have CNS effects unrelated to seizures, maximal electroshock (MES) was chosen as the priming seizure. Flurothyl was the second convulsant agent.

In our preliminary studies¹ it was determined that donor-CSF collected from the cisternal space of rats at various times post-convulsion increased the threshold to convulsion in the recipient animals. If the recipients were pretreated with an opioid antagonist the anticonvulsant activity of the EAS was blocked. Thus, it appeared that an EAS, opioid in nature, was present in the donor-CSF.

Subsequent experiments² have revealed the following biochemical and pharmacological properties of this EAS. Its bioactivity is not affected when exposed to the specific enkephalinase inhibitors thiorphan and bestatin, yet its anticonvulsant bioactivity is dramatically increased in the presence of the peptidase inhibitors aprotinin and bacitracin, and completely degraded when heated (90°C) or exposed to immobilized trypsin. Ultrafiltration of the donor-CSF revealed that the EAS was associated with bioactive material in the 5000-10000 molecular weight range. The immunoreactive profile of the donor-CSF supports the results of the ultrafiltration experiments in that only β -endorphin-like immunoreactive material and a large [Met]enkephalin precursor molecule are present. Preliminary molecular weight sizing experiments using gel exclusion chromatography suggest that the major species in the donor-CSF yielding this [Met]enkephalin-like immunoreactivity migrates with a molecular mass of 8700 Da (D. Liston, pers. commun.).

Direct evidence for an opioid comes from *in-vivo* studies where the EAS was found to be highly sensitive to both naloxone and the selective δ -opioid antagonist ICI-174864³. More recently, Porreca and Davis at the University of Arizona have determined that the EAS produces naloxone-reversible, concentration-dependent inhibitions of the isolated mouse vas deferens and volume-induced micturition contractions (bladder motility) in rats. Collectively, these characteristics suggest that the EAS may be a large molecular weight opioid peptide.

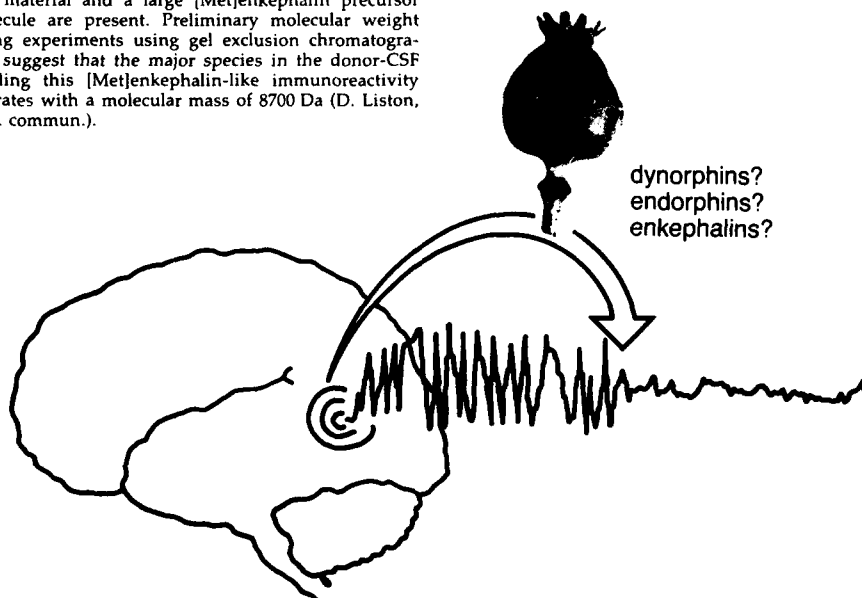
A potential candidate for a seizure-activated EAS might be expected to satisfy certain biological criteria such as:

- exogenous administration should provide seizure protection
- this 'anticonvulsant' effect should represent a dose-related, pharmacological (receptor-mediated) action of the neuromodulator
- endogenous seizure-related changes in levels, receptors, etc. should be measurable
- administration of specific antagonists of the EAS should attenuate postseizure inhibition
- since the EAS is not tonically active, specific antagonists would not be expected to influence pre-seizure or seizure activity
- it would be predicted that the putative EAS candidate would be localized within the CNS, most likely in areas believed important to the modulation and control of epileptic activity, functionally hyperpolarizing, rather than depolarizing, central neurons.

At the moment, endogenous opioid peptides appear to most fully satisfy these criteria.

References

- 1 Tortella, F. C. and Long, J. B. (1985) *Science* 228, 1106-1108
- 2 Tortella, F. C. and Long, J. B. (1988) *Brain Res.* 456, 139-146



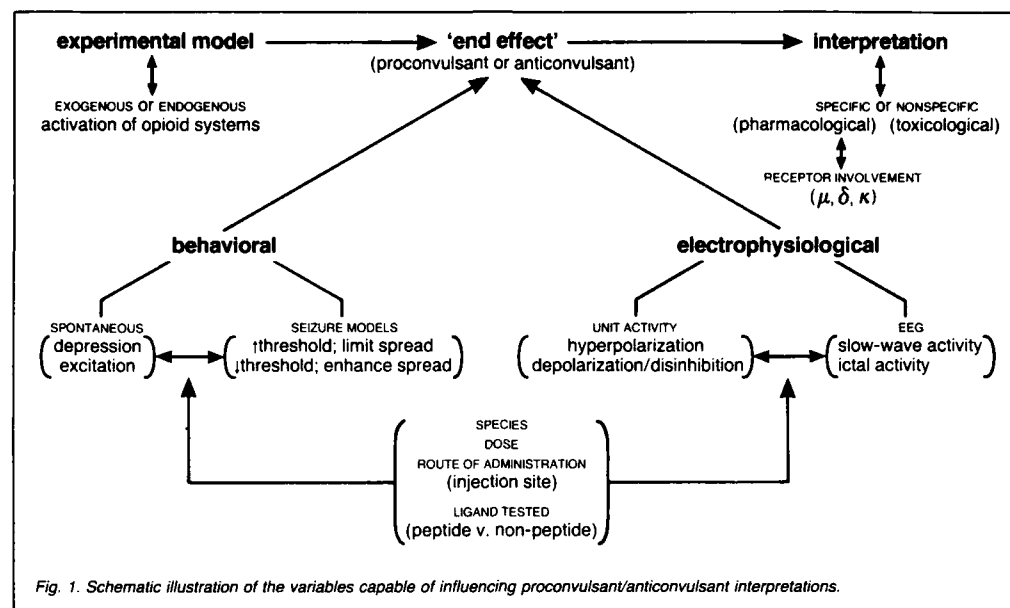


Fig. 1. Schematic illustration of the variables capable of influencing proconvulsant/anticonvulsant interpretations.

modulate postictal inhibition⁵, the inherent ability of a seizure to inhibit (or self-limit) the recurrence of subsequent seizure episodes.

In 1982 we used a two-seizure rat model to demonstrate that an initial MES convulsion increased the seizure threshold to a subsequent flurothyl convulsion, an effect significantly attenuated by naloxone but not by the GABA antagonist bicuculline³³. These findings were later reinforced when the effects of repeated MES convulsions on the progression of seizure severity were examined. Here, the generalized convulsions produced by a series of six intermittent MES treatments (administered at 10 min intervals) were observed to progressively decline in terms of severity and duration. The protective effects associated with the repeated seizures could be stereospecifically reversed with naloxone and prevented by morphine tolerance or hypophysectomy⁵. Collectively, these results provided evidence for endogenous opioid involvement in the mechanisms of postseizure inhibition.

That endogenous opioids may play a role as inhibitory neuromodulators of seizure arrest and refractoriness has been addressed in several other studies. The anticonvulsant effects of

electroconvulsive shock against kindled seizures can be significantly reduced by naloxone, as can the postseizure inhibition associated with massed kindling trials. The postictal inhibition associated with the spontaneous seizures of the genetically predisposed epileptic gerbil has also been reported to be attenuated by naltrexone.

In all of these studies (see Refs 5, 22 and 34), including the repeated MES model, moderate to high doses of naloxone (1–10 mg kg⁻¹) were required to effect the post-seizure inhibition. While these doses are excessive for pharmacological antagonism it is difficult to make direct comparisons, since under these experimental conditions the actual concentration of endogenous ligand at the receptor is unknown.

Interestingly, studies measuring changes in opioid receptors following seizures have demonstrated increases in δ -opioid receptor dynamics with an apparent lack of plasticity in μ -opioid recognition sites^{35–37}. In opioid pharmacology it is well established that while μ -receptor-mediated responses are highly sensitive to naloxone (microgram doses), the lower affinity of naloxone for non- μ -opioid receptors dictates that δ -mediated actions, usually require *in-vivo* doses of 1–10 mg kg⁻¹ for inhibition. There-

fore, as with all other opioid-like postictal effects of seizures^{5,27}, the postseizure inhibition described in these studies may be primarily influenced by an endogenous δ -opioid receptor system. Defining precisely which opioid receptor(s) modulate postseizure inhibition awaits thorough studies using selective antagonists specific for either μ - (e.g. β -fNA), δ - (e.g. ICI-174864) or κ - (i.e. nor-BNI) opioid binding sites.

□ □ □

It is becoming increasingly evident that many of the divergent conclusions, and at times conflicting interpretations, concerning opioid peptides and epilepsy may be due to the syndrome of 'failing to see the forest for the trees'. Conclusions regarding opioid involvement in epilepsy are too often made on the basis of only one experimental approach in a single species or test system, without consideration of pharmacological specificity for the opioid system. This review was not intended to provide the 'Rosetta Stone' for understanding endogenous opioids and epilepsy. However, when the experimental efforts of the past ten years are considered collectively it is obvious that the effective balance of opioid peptides to enhance or depress CNS excitability

involves interrelated mechanisms, the outcome of which depends upon many experimental variables (Fig. 1).

Composite behavioral, electrophysiological, anatomical and biochemical evidence implies an important functional role for opioid peptides in the mechanisms of postseizure arrest and refractoriness. Perhaps the most important application of this hypothesis lies with our understanding of the neuronal events mediating the interictal transition from single seizures to the recurrent attacks. Can we consider that the focal and generalized depolarization associated with abnormally discharging epileptic neurons activates an endogenous opioid anticonvulsant substance which, in concert with other putative neuromodulators of endogenous anticonvulsant activity, functions to quiet the 'seizing' brain? Perturbation of such a system, either genetically or otherwise, could explain the occurrence in epilepsy of the subpopulation of patients with status epilepticus, or the pathological transition from a 'self-limited' epileptic attack to this life-threatening medical emergency.

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U-50488: 3,4-dichloro-N-[2-(1-pyrrolidinyl)-cyclohexyl]benzeneacetamide
 nor-BNI: nor-binaltorphimine
 ICI-174864: Allyl-Tyr-Aib-Phe-Leu-OH
 ICI-154129: N,N-bisallyl-Tyr-Gly-Gly-ψ-(CH₂S)-Phe-Leu-OH
 α-NEO: α-neo-endorphin
 DAME: [D-Ala², Met⁵]enkephalinamide
 DADL: [D-Ala², D-Leu⁵]enkephalin
 PL017: [N-MePhe³, D-Pro⁴]morphiceptin
 DAMGO: [D-Ala², N-MePhe⁴, Gly-ol⁵]enkephalin
 DSTLE: [D-Ser², Leu⁵, Thr⁶]enkephalin
 DPDPE: [D-Pen², D-Pen⁵]enkephalin
 E₂₅: Tyr-D-Ala-Gly-Phe-D-Leu-OMe
 FK33-824: [Tyr-D-Ala-Gly-N-Me-Phe-Met⁵]ol
 CTP: D-Phe-Cis-Tyr-D-Trp-Lys-Thr-Pen-Thr-NH₂